Median Lethal Concentration and Efficacy of *Bacillus Thuringiensis* Against Banded Sunflower Moth (Lepidoptera: Tortricidae)

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ABSTRACT This study was conducted to determine dose-mortality response of the banded sunflower moth, *Cochylis hospes* Walsingham, to *Bacillus thuringiensis* Berliner and its effect on sunflower seed damage and yield. Of *B. thuringiensis* products tested, Cutlass AF and Javelin WG had the lowest LC_{50} values and highest slopes and relative potencies for *C. hospes.* Javelin WG was superior to the other *B. thuringiensis* materials tested and to Asana XL in preventing seed damage. Sunflower heads sprayed with Javelin WG had higher yields than sunflower heads sprayed with Asana XL or the other *B. thuringiensis* products tested. Asana XL is a standard chemical insecticide and is considered to give good control of banded sunflower moth. As an alternative to Asana XL, any of the *B. thuringiensis* products tested could be employed for control of *C. hospes* larvae and to reduce the impact on beneficials in sunflower fields.

KEY WORDS *Cochylis hospes, Helianthus annuus, Bacillus thuringiensis*

THE BANDED SUNFLOWER MOTH, *Cochylis hospes* Walsingham, is an important economic pest of cultivated sunflower, *Helianthus annuus* L., in North America (Charlet et al. 1997). Early instars feed primarily on sunflower pollen and floral tissues, and 3rd and later instars cause most of the economic damage by feeding on seeds (Rogers 1978, Charlet and Gross 1990). Although insecticides are commonly used for sunflower insect pest management, their use can adversely affect natural enemies and pollinating insects (Rogers and Kreitner 1983). *Bacillus thuringiensis* Berliner variety *kurstaki* is a possible alternative to synthetic insecticides for controlling lepidopterous pests on sunflower. It is safe for predators and other nontarget insects (Lüthy et al. 1982) and does not cause environmental pollution. *B. thuringiensis* has been successfully used to control lepidopterous pests on a variety of crops (Falcon 1971, pp. 67–90), including sunflower moth, *Homoeosoma electellum* (Hulst) (Brewer 1991).

Reynolds (1960) urged the development of baseline dose-mortality curves for promising insecticides against pest species of significant economic importance before those species develop insecticide resistance. To determine median lethal concentrations (LC_{50}) of an insecticide against a susceptible insect population, probit units of percent mortality are customarily plotted against a logarithmic scale of dosages. This approach was first proposed by Bliss (1935).

The purpose of this study was to determine the median lethal concentration values (LC_{50}) of 7 *B. thuringiensis* products and to evaluate their efficacy compared with a chemical insecticide registered for control of the banded sunflower moth.

Materials and Methods

Bioassay Experiments. Banded sunflower moth larvae were obtained from a colony maintained in the USDA-ARS Bioscience Research Laboratory, Fargo, ND. The rearing procedure used to maintain the parental stock culture in the laboratory was described by Barker (1988). Seven commercial formulations of *B. thuringiensis* were tested. See Table 1 for the products and concentrations used.

The dose-mortality response of each *B. thuringiensis* formulation was tested by topically applying 150-μl aliquots of each concentration to the surface of artificial diet in 29.6 ml cups. Controls were treated with 150 μl of distilled water. The solution was spread evenly over the diet surface (600 mm^2). Diet was prepared as described by Vanderzant et al. (1962) and modified by Barker (1988). After the treated diet had air-dried, a single 1-d-old larva was placed in the center of each cup. Each concentration and the control were tested using 30 larvae. After the cups were closed, they were placed into plastic trays and held in a growth chamber at 28.5 ± 1°C and a photoperiod of 14:10 (L:D) h. Larvae were allowed to feed on the treated diet for 72 h. On day 3, larval mortality was recorded. The complete series of tests were done twice.

Field Experiments. Field trials were conducted at the North Dakota State University Research Farm Site, near Prosper, ND, during the sunflower growing seasons of 1995 and 1996. A four-row mechanical planter was used to plant sunflower hybrid '894' during the 4th wk of May. Plots were 4 rows wide by 10 m long, with 76.2 cm between rows. The plants were thinned at the two-leaf stage to ≈25–30 cm apart

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Table 1. *B. thuringiensis* products tested and concentrations used

B. thuringiensis products	Concentrations, $\mu g/\mu l$
Dipel 2X WP (wettable powder, Abbott, North Chicago, IL)	0.67×10^{-2}, 0.61×10^{-3}, 0.16×10^{-3}, 0.55×10^{-4}, 0.21×10^{-4}, 0.81×10^{-5}
Javelin WG (wettable granule, Sandoz, Des Plaines, IL)	0.67×10^{-2}, 0.61×10^{-3}, 0.16×10^{-3}, 0.55×10^{-4}, 0.21×10^{-4}, 0.81×10^{-5}
Xen Tari (water dispensable granule, Abbott, Des Plaines, IL)	0.1×10^{1}, 0.91×10^{-3}, 0.24×10^{-3}, 0.83×10^{-4}, 0.31×10^{-4}, 0.12×10^{-4}
Cutlass WP (wettable powder, Ecogen, Langhorne, PA)	0.87×10^{-2}, 0.79×10^{-3}, 0.21×10^{-3}, 0.72×10^{-4}, 0.27×10^{-4}, 0.11×10^{-4}
Thuricide AF (aqueous flowable, Voluntary, Bonham, TX)	0.53×10^{1}, 0.48×10^{-2}, 0.13×10^{-2}, 0.44×10^{-3}, 0.17×10^{-3}, 0.64×10^{-4}
Condor OF (flowable, Ecogen, Langhorne, PA)	0.87×10^{-2}, 0.79×10^{-3}, 0.21×10^{-3}, 0.72×10^{-4}, 0.27×10^{-4}, 0.11×10^{-4}
Cutlass AF (aqueous flowable, Ecogen, Langhorne, PA)	0.21×10^{-3}, 0.19×10^{-2}, 0.52×10^{-3}, 0.18×10^{-3}, 0.67×10^{-4}, 0.26×10^{-4}

within the rows. The plots were equivalent to 47,000 plants per hectare. There was a 2.5-m fallow area between plots and 3.5-m fallow areas between blocks. Treatments were arranged in a randomized complete block design with 4 replications. Standard cultivation practices for this location were used to maintain the plants. When sunflower capitula were at bloom stage (R5.1–5.3, Schneiter and Miller 1981), insecticide treatments were applied using a tractor-mounted boom sprayer (40 psi, S-11004 nozzles, 1,700 rpm, 117 liters of H_2O/ha).

At physiological maturity (R9), sunflower heads from 5.3 m of the 2 center rows of each plot were hand-harvested, oven-dried, hand-threshed, and the seeds were hand-cleaned by manually removing debris. Ten samples (100 seed each) were selected randomly from each plot and were evaluated for the percentage of banded sunflower moth damaged seeds. In addition, 10 samples (100 seed each) were selected randomly from each of the 4 replicates and weighed to determine seed yields per treatment.

Statistical Analysis. Dose-mortality response data from the bioassays were tested by probit analysis (LeOra Software 1987). POLO-PC uses Abbott's formula to incorporate control mortality (Abbott 1925). The chi-square value was used to measure the goodness-of-fit of the probit regression line to the points. PROC univariate, residual analysis was used to check if the variables for the field data met assumptions of analysis of variance. Data were analyzed using the general linear model (SAS Institute 1995), and means were compared by using least significance difference least significant difference ($P < 0.05$).

Results and Discussion

Bioassay Experiments. Banded sunflower moth dose-mortality responses to the tested *B. thuringiensis* products are summarized in Table 2. The dose-mortality relationship of an insect to a toxin is typically expressed as an LC_{50} value, which is the toxin concentration required to kill 50% of the population in a specified period. The lower the LC_{50} value, the greater the toxicity. Cutlass AF and Javelin WG (wettable granules) had the lowest LC_{50} values, suggesting that they were more toxic to *C. hospes* larvae than Cutlass WP (wettable powder, Thuricide AF, and Condor OF with the highest LC_{50} values. However, the LC_{50} alone does not reveal an accurate picture of the total pathogenic effect. Insects respond to increasing doses of pathogenic organisms by increased infection and mortality, just as they respond to increased doses of insecticides (Metcalf and Luckman 1994). Therefore, the mortality effect of an insect pathogen on its host can be expressed as an LC_{50} value and can also be characterized by the slope of the log-probit curve.

The slope of the dose-mortality curve is a measure of variability in response to treatment within the insect population tested. As the value of the slope increases, mortality associated with changes in concentration increases. Conversely, as the value of slope decreases, less change in mortality is seen per unit change in concentration of the mortality agent. Insect pathogens, such as polyhedrosis viruses, have slopes of 0.86 or less, whereas insecticides, such as DDT, have slopes of 5.5 or greater (Metcalf and Luckman 1994). Insect pathogens that produce toxins, such as *B. thuringiensis,* are usually characterized by intermediate slopes of ≈2.58 (Bucher 1960, Burges and Thomson 1971). The slopes of the dose-mortality curves for Cutlass AF and Javelin WG were 2.02 and 1.07, respectively, suggesting that these products have some toxicity to the banded sunflower moth and do not rely, at least solely, on infection to cause mortality.

Relative potencies are used to compare the degree of effectiveness of various *B. thuringiensis* materials against a standard. We compared all the *B. thuringien-*

Table 2. Slopes, median lethal concentration (LC_{50}) values with fiducial limits (FL), and relative potency of *B. thuringiensis* products against 1st-instar *C. hospes* larvae ($n = 60$)

B. thuringiensis products	Slope ± SE	LC_{50} (95% FL), $\mu g/\mu l$	Relative potency
Dipel 2X WP	1.0572 ± 0.1565	0.29×10^{-3} ($0.14 \times 10^{-3} - 0.58 \times 10^{-3}$)	1.000
Javelin WG	1.0789 ± 0.1190	0.08×10^{-3} ($0.04 \times 10^{-3} - 0.14 \times 10^{-3}$)	3.748
Xen Tari	1.1849 ± 0.1312	0.18×10^{-3} ($0.11 \times 10^{-3} - 0.29 \times 10^{-3}$)	1.562
Cutlass WP	0.7506 ± 0.1024	0.53×10^{-3} ($0.10 \times 10^{-3} - 0.92 \times 10^{-3}$)	0.636
Thuricide AF	1.1079 ± 0.1210	0.42×10^{-3} ($0.23 \times 10^{-3} - 0.76 \times 10^{-3}$)	0.694
Condor OF	0.6388 ± 0.1622	0.33×10^{-3} ($0.16 \times 10^{-3} - 0.67 \times 10^{-3}$)	0.882
Cutlass AF	2.0259 ± 0.2550	0.06×10^{-3} ($0.03 \times 10^{-3} - 0.11 \times 10^{-3}$)	4.976

Table 3. Comparison of an insecticide and several commercial *B. thuringiensis* products tested against banded sunflower moth larvae for percent of damaged seed, seed weight, and seed yield in sunflower, Prosper, ND

Insecticides	Application rate (kg/ha)	% damaged seeds		1,000 seed wt, g		Seed yield (g/0.0025 ha)	
		1995	1996	1995	1996	1995	1996
Dipel 2X WP	1.1 kg/ha	9.1 ± 1.6b	3.5 ± 0.3d	36.2 ± 0.2a	51.5 ± 1.7a	999.1 ± 65.7a	1,908.5 ± 135.9a
Javelin WG	1.7 kg/ha	7.2 ± 1.2b	3.1 ± 0.4d	37.1 ± 0.1a	52.6 ± 3.4a	1,217.9 ± 39.3a	2,403.2 ± 338.7a
Xen Tari TM	1.1 kg/ha	8.6 ± 0.9b	5.8 ± 0.5bc	36.0 ± 0.2a	54.0 ± 3.2a	1,258.6 ± 31.6a	1,912.2 ± 97.5a
Thuricide AF	4.2 liters/ha	7.6 ± 1.0b	5.7 ± 0.4bc	36.7 ± 0.2a	50.1 ± 3.0a	1,124.8 ± 39.6a	1,867.7 ± 100.7a
Condor OF	3.8 liters/ha	7.3 ± 1.0b	7.3 ± 1.1b	33.3 ± 0.1a	49.9 ± 1.5a	1,055.3 ± 24.3a	1,825.7 ± 114.9a
Asana XL	0.4 liters/ha	7.1 ± 1.3b	4.6 ± 0.4cd	35.1 ± 0.2a	51.9 ± 2.5a	1,185.5 ± 93.6a	1,926.2 ± 172.8a
Control	–	14.3 ± 0.8a	13.8 ± 0.3a	39.5 ± 0.2a	53.1 ± 2.6a	1,172.5 ± 99.7a	1,688.7 ± 207.1a

Column means followed by the same letter are not significantly different ($P \leq 0.05$, LSD).

sis formulations against Dipel WP 2X. A relative potency >1 indicates the product was more toxic to 1st-instar banded sunflower moth larvae than was Dipel WP 2X. Cutlass AF and Javelin WG had the highest relative potencies and were the most efficacious materials tested against *C. hospes* larvae.

Field Experiments. In general, the microbial and chemical insecticides were equally effective in limiting seed damage (Table 3). The one exception was sunflower treated with Condor OF in 1996, which had significantly more seed damage than the insecticide check, Asana XL. Insecticide treatments did not result in significant differences in seed weight and seed yields (Table 3) compared with controls for either year. This may be caused by the relatively low infestation levels both years. However, in 1996, but not 1995, Javelin WG had higher, although not significantly higher, yields than either Asana XL or the other *B. thuringiensis* products tested. Asana XL, a chemical insecticide, is a standard insecticide used on sunflower and is considered to give good control of banded sunflower moth. Although as an alternative to Asana XL, any of the *B. thuringiensis* insecticides tested could be used, those with lowest LC_{50} values, and highest slopes and relative potencies would probably be most effective. The additional benefit from using *B. thuringiensis* is the absence of any adverse impact on natural enemies and insect pollinators.

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